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Effect of Handling Procedures on Sample Quality

Arthur D. Little, Inc., Cambridge, Mass.

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EFFECT OF HANDLING PROCEDURES ON SAMPLE QUALITY

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16. ABSTRACT The report gives results of an evaluation of the effects of typical shipping and storage handling procedures on organic materials collected in Level 1 environmental assessment (EA) studies. Parameters reviewed included: sample container composition (amber glass and high-density linear polyethylene), head space composition (air or nitrogen), temperature (38 C, 21 C, and 5 C), lighting (dark and diffuse sunlight), and catalytic species content. Three sample sets, representing fractions obtained during a Level 1 EA and containing six model organic compounds, were used. A simulated 3-week shipping and storage cycle represented elapsed time between sample collection and analysis. All three experiments were in accordance with statistical principles appropriate for conducting factorial experiments. Experimental results were analyzed using analysis of variance, to assess the relative effect of each shipping/storage condition studied.

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SUMMARY

The results of a study designed to evaluate the effects of typical shipping and storage handling procedures on organic materials collected in Level 1 studies are presented. Specific parameters reviewed included sample container composition (amber glass and high density linear polyethylene), head space composition (air or nitrogen), temperature (38°C, 21°C, 5°C), lighting condition (dark and diffuse sunlight) and catalytic species content. Three unique sample sets, representative of fractions obtained during a Level 1 environmental assessment, were formulated containing six model organic compounds. A three-week simulated shipping and storage cycle was used as representative of elapsed time between sample collection and analysis. All three experiments were designed in accordance with statistical principles appropriate for conducting factorial experiments. Experimental results were analyzed by the Analysis of Variance technique in order to assess the relative effect of each shipping/storage condition studied.

The major findings of these experiments are as follows:

- Generally, any reasonable combination of shipment temperature, head space composition, and storage condition can be utilized for the short term storage of Level 1 survey analysis samples.

- These same conditions can be employed for the short term storage of samples undergoing quantitative chemical analysis where the accepted range of variation is $\pm$ 10 percent.

- Amber glass containers are preferred for sample storage over polyethylene bottles, as it has been found that spurious contaminants may be extracted from the container walls by contact with organic solvents.

- Diffuse sunlight can penetrate polyethylene containers and cause degradation in species that are photoreactive. Amber glass on the other hand will absorb the harmful wavelengths prohibiting photodegradation.

I. INTRODUCTION

Sampling is conducted as part of a Level 1 environmental assessment study which provides data on the overall composition of process and effluent streams. The collected samples constitute a small portion of the stream and are assumed to be representative of the entire stream. Therefore, the immediate goal of any process involving sample collection is to insure that the results obtained from that portion are truly representative of the whole. As such, all sampling and analysis schemes are developed using best state of the art procedures in an attempt to minimize the occurrence of uncharacteristic changes. If interferences are encountered, appropriate steps are implemented to either quantify their contribution or to eliminate them altogether. Failing this, the methods are rejected and alternative procedures explored.

Despite the rigorous method development practices, there still are areas where changes can occur and not be detected. One such instance occurs between the time a sample is first placed within a container, and when it is removed for analysis. During this time, the sample may be exposed to a number of adverse conditions, such as heat or light, that can result in change or degradation.

Normally, it is preferable to minimize the length of time that any sample is retained prior to analysis, as this reduces the available time for changes to occur. However, in many instances, this cannot be done, and alternative precautionary preservation procedures must be devised to control sample quality.

Three factors are of concern in the development of any preservation technique: contamination, loss and chemical reactivity. Any one or combination of these factors can happen during the shipment and storage of samples, compromising the value of their analysis. It is important, therefore, that practices be designed to alleviate, if not eliminate, their occurrence.

Many handling procedures are documented within the literature, all of which are designed to fulfill a specific requirement. For example, plastics of any sort (excluding Teflon) have been shown to adsorb sample constituents or introduce extraneous contaminants (1, 2). Specific classes of contaminants introduced include plasticizers, antioxidants, colorants and stabilizers (3). Thus, many researchers indicate a preference for glass containers, equipped with Teflon or aluminum foil cap inserts (1, 2, 4). However, even this technique is not foolproof as both Teflon and aluminum may be contaminated with up to 400 parts per billion of di-2-ethylhexyl phthalate (5).

Elaborate container and apparatus cleaning procedures are outlined in a number of references (6, 7, 8). Coupled with extensive pre-utilization container storage specifications, these procedures minimize spurious contamination but do not completely eliminate their possible occurrence.

During actual shipment and storage operations, many researchers (1,7,9,10) have noted preferences for either sub-zero temperature conditions or freeze drying of sample media, but even these precautions do not eliminate possible compositional variations over extended periods of time.

To further complicate matters, many desirable handling practices cannot be implemented and rigorously followed under adverse field collection conditions. Assuming that the analyst is able to collect samples and package them according to defined practices, their destinations and environments are not easily regulated unless they are always under the immediate control of the investigator during shipment and storage.

Recently, the Process Measurements Branch of the Industrial Environmental Research Laboratory (IERL) published a manual detailing specific procedures to be used for the collection and analysis of materials during a Level 1 environmental assessment. However, since little quantitative data was available describing the impact of handling procedures, few recommendations were presented for proper shipment and storage practices. To supplement the manual, the Process Measurements Branch of the IERL commissioned this study to review normally encountered procedures and provide data describing the impact of chosen conditions upon the sample quality of organic species in the samples.

Although a complete review of methods is impractical, it is hoped that sufficient data will be produced to highlight practices that may be practically implemented and result in minimum sample degradation and contamination.

II. EXPERIMENTAL CONSIDERATIONS

A. General Considerations

Three main factors contribute to the degradation of sample quality: contamination, sample loss, and chemical reactivity or instability. Given appropriate conditions, any one or combination of these factors may occur and become a source of interference within the retained sample, detracting from its representative nature. A number of alternative shipment and storage procedures have been proposed and implemented, many of which include provisions for the regulation of at least one of the following parameters.

- shipping and storage time
- sample container composition and quality
- shipment and storage temperature
- container head space composition
- storage lighting condition
- presence of catalytic species

Little quantitative information is available detailing the potential impact of these three factors upon mixed samples. Within this study, various levels of each of these parameters were experimentally controlled to a assess their contribution to degradation within samples.

It was decided at the outset that test samples would be developed which were representative of the type of sample collected during a Level 1 environmental assessment and which contained a variety of classes of compounds. These representative samples would be packaged, stored under simulated shipping conditions and returned to the laboratory where they would await analysis. The simulated shipment time would span one week and the laboratory storage time an additional two weeks. Analyses would be performed within one week of the termination of the storage period.

B. Experimental Design

Using criteria presented in the Level 1 Procedures Manual, three experiments were designed to study the effects of the following parameters, or factors, on sample integrity:

- sample container composition (amber glass or polyethylene),
- head space (air, nitrogen, or a combination),
- shipment and storage temperatures (38°C, 21°C, 5°C),

- storage lighting conditions (dark or diffuse sun), and
- catalytic species content (acid extractable stainless steel components).

Sets of samples were devised to simulate representative probe wash (Set I), sorbent trap condensate extract (Set II), and sorbent resin samples (Set III) which are obtained during a Level 1 source assessment. Once formulated and packaged, each sample was handled according to practices normally encountered during transport and storage cycles.

Some combinations were eliminated as non-applicable, such as the conversion back to an air head space over samples after preliminary packaging under nitrogen. Furthermore, a variation in catalytic species content was considered only in the Set II (condensate extract) experiment. The sorbent resin experiment (Set III) was conducted with additional limitations in the factor/level combinations tested. However, some of the factor/level combinations were repeated in order to achieve greater precision in estimating experimental error. The final array of factors and levels chosen for the three experimental sets is given in Table 1.

As the experimental program progressed it was established that certain economies in experimentation were possible by employing statistical principles of experimental design. Therefore, only twenty-seven unique combinations were actually tested in the condensate extract experiment (Set II). However, the set of twenty-seven input conditions were selected according to the theory of fractional factorial experimentation (11, 12), so that meaningful inferences could be made about each factor independently, as well as certain predetermined two-factor interactions. These concepts are described in greater detail in Section IV-A.

C. Sample Identification

A coding system was developed that would allow for the immediate identification of any sample without reference to a master log. The code was derived from the first letter of each important shipping or storage condition; an example of which is given below:

Container	Head Space Composition	Shipment Temperature	Storage Condition	Catalytic Species Content
AG-	NN-	H-	CD-	S
Packaged in an amber glass bottle	Shipped in N_2 Stored in N_2	Shipped under elevated temperature (hot)	Stored in the cold and in the dark	Contains acid extractable stainless steel constituents

A complete list of sample coding abbreviations is presented in Table 1.

TABLE 1

Admissible Experimental Conditions

FACTORS	Code	LEVELS: Probe Wash Experiment (Set I)	Condensate Extract Experiment (Set II)	Sorbent Resin Experiment (Set III)
(1) Container	AG	Amber Glass	Amber Glass	Amber Glass
	P	Polyethylene	Polyethylene	(Not analyzed)
(2) Shipment/Storage Head Space Composition	AA	Air/Air	Air/Air	Air/Air
	AN	Air/Nitrogen	Air/Nitrogen	(Not analyzed)
	NN	Nitrogen/Nitrogen	Nitrogen/Nitrogen	Nitrogen/Nitrogen
(3) Shipment Temperature	H	Hot (37°C)	Hot	Hot
	R	Room (21°C)	Room	Room
	C	Cold (5°C)	Cold	Cold
(4) Storage Condition Temperature/Lighting	RD	Room/Dark	Room/Dark	Room/Dark
	RL	Room/Light	Room/Light	(Not analyzed)
	CD	Cold/Dark	Cold/Dark	Cold/Dark
(5) Catalytic Species	N	Not analyzed	None	Not analyzed
	S		Stainless Steel Components	
Possible Combinations		$3^3 \times 2 = 54$	$3^3 \times 2^2 = 108$	$3 \times 2^2 = 12$

D. Sample Formulation

Model compounds selected for review within this study were chosen on the basis of the following criteria:

- Representative of a variety of different compound classes.
- Representative of a boiling point range where losses due to volatility may be possible, but do not pose a major influence.
- Included in concurrent Level 1 analysis studies.
- Applicability to ready analysis by gas chromatography.

Based on these selection criteria, the following six compounds were subsequently used:

Phenol
N-Methylaniline
4-Chlorobenzaldehyde
Acenaphthene
n-Hexadecane
4,4'-Dichlorobiphenyl

Specific structural and physical parameters of the chosen species are detailed in Table 2.

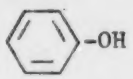
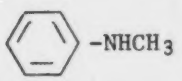
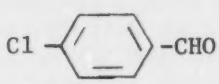
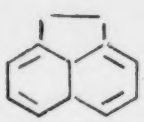
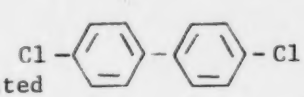
Using information detailed within the "Level 1 Procedures Manual" (6), three sets of simulated field samples were formulated, namely probe wash (Set I), sorbent trap condensate extract (Set II), and sorbent resin (Set III). Each set was compounded such that typically expected field concentrations of selected model species were contained within the individual sample. To minimize the variability of compound concentration within any given sample, master batches of each sample type were premixed and aliquots removed for packaging.

As compounded, the probe wash and condensate extract series contained between 30 and 50 milligrams of each model compound, diluted to a final volume of 300 milliliters with carrier solvent. As outlined in the Procedures Manual, the probe wash (Set I) solvent was 1:1 methylene chloride: methanol, and the condensate extract (Set II) solvent was methylene chloride. The sorbent resin samples contained approximately 3 to 5 milligrams of each model compound on 15 ±1 grams of XAD-2. In this instance a master batch of spiking solution was made up in pentane and 5 mls pipetted onto the resin.

During formulation of the condensate extract set, an attempt was made to simulate conditions encountered during field utilization of a SASS train. It was reported that sorbent trap condensate samples obtained from sources having hydrochloric acid mist and sulfur dioxide within the effluent stream, contained quantities of hydrochloric and

TABLE 2

Model Compound Chemical and Physical Properties

Species	Class	Structure	Molecular Weight	Boiling Point °C
Phenol	Oxygenated compound (phenol)		94	182
N-Methyl aniline	Nitrogen compound		107	196
4-Chlorobenz-aldehyde	Oxygenated compound (Aldehyde)		141	214
Acenaphthene	Polynuclear Aromatic Hydrocarbon		154	277
n-Hexadecane	Straight Chain Hydrocarbon	$CH_3(CH_2)_{14}CH_3$	226	287
4,4'Dichloro-biphenyl	Halogenated Compound (Polychlorinated biphenyl)		223	315

sulfuric acid and exhibited a characteristic pale green color (13). As the sorbent trap is fabricated of stainless steel, the color possibly results from the etching of the metal by the acid and is indicative of trace metallic content within the condensate liquor. To approximate this condition, and assess its potential impact upon storage stability parameters, simulated green and white liquor solutions were formulated and extracted with methylene chloride for Set II samples. Specific details of this process and all other sample formulation operations are provided within the appendices of this report. (Tables A1-A3)

E. Handling Cycle

After formulation, samples were placed within the desired container type, with either a nitrogen or air head space, and sealed. They then were subjected to a three-week handling cycle. During the first week, or simulated shipment phase, selected samples were stored within closed boxes and placed in controlled temperature zones to simulate conditions expected to arise during truck transport from a remote collection point to a central laboratory. Three such zones were used: one of 38°C (100°F), one of 21°C (70°F), and one of 5°C (40°F). All samples were then unpacked and rerouted for two additional weeks to a storage location. Samples requiring head space composition change were opened, purged with nitrogen and then resealed before being stored. During the storage phase, three basic combinations existed:

21°C (70°F) and	diffuse sunlight (northern window)
21°C (70°F) and	dark (closed cabinet)
5°C (40°F) and	dark (closed refrigerator)

F. Analytical Procedures

Gas chromatography (GC) was utilized as the major quantitative analysis tool during this evaluation. Specific experimental details are presented in the appendices of this report (Table C-1). As the probe wash and condensate extract samples were already dilute solutions, no sample preparation was required prior to analysis. For sorbent resin samples, a continuous 24-hour Soxhlet extraction was required to remove the spiked compounds from the XAD-2 resin. Since the sample recovery of this step was unknown, several non-stored spikes were similarly prepared and analyzed along with the stored samples.

This was accomplished by placing 15 $\pm$1 gm of XAD-2 resin into an amber glass bottle, and spiking it with 5 mls of a freshly prepared standard solution. The contents of the bottle were then immediately transferred into a Soxhlet thimble and extracted for 24-hours with roughly 200 milliliters of methylene chloride. The resulting solution was brought to a final volume of 250 milliliters with methylene chloride, from which a 5 microliter aliquot was removed

and directly analyzed by GC. Additional details of this procedure are discussed in Section III of this report.

In all cases, a fresh standard solution was prepared immediately before the analysis of individual sample sets. This standard was required for several reasons. First, this sample was useful for monitoring the instrumental parameters. If integrated peak area responses were found to change between repetitive injections of the standard solution, steps could be taken to retune the GC to original conditions.

Similarly, as it was impractical to analyze all stored samples on the same day, the fresh standard provided a mechanism through which the data from non-contiguous analysis could be compared. At the beginning of each day, the standard solution was injected several times to insure that the instrumental response was equivalent to prior repetitions. If not, all parameters were rechecked and adjusted, prior to proceeding with the analysis of the stored samples.

Finally, this sample served as a calibtation standard. Formulated to duplicate the composition of the stored samples, when they were originally packaged and placed in the simulated shipping sequence, this standard solution represented the stored samples in an unaltered state. By directly comparing the observed peak area responses of the stored sample to the fresh standard, and correcting for minor initial compounding variations, it becomes possible to compute a percentage deviation attributable to the imposed handling procedures.

The metallic content of the green and white liquors and the methylene chloride extract of the green liquor was determined by spark emission spectroscopy (ES). Attempts to obtain more accurate quantitative data for the methylene chloride extract by the inductively coupled plasma optical emission spectroscopy (ICPOES) technique was frustrated by the lack of compatibility between this technique and methylene chloride. However, quantitative results were obtained for both of the aqueous phase solutions (green and white liquor) by ICPOES. All results of these analyses are given in Appendix B, Tables B-1 and B-2.

G. Data Analysis and Interpretation

The extent of sample degradation is computed as the ratio of the stored samples' area response to that of the freshly prepared standards. For example, in the hypothetical situation where a sample initially containing 100 µg/ml of phenol yielded an integrated area of 500 microvolts-seconds and a standard containing 105 µg/ml phenol exhibian area response of 515 microvolts-seconds, the equation would be

$$\frac{500 \ \frac{\text{microvolts-seconds}}{\text{sample}}}{515 \ \frac{\text{microvolts-seconds}}{\text{standard}}} \times \frac{105 \ \mu g/ml}{100 \ \mu g/ml} = 1.019$$

A value of 1.00, representing the expected concentration, is subtracted from this value and the resulting decimal expressed as a percentage, i.e.,

$$1.019 - 1.00 = 0.019 = 1.9\%$$

This figure is the percentage deviation of the stored samples' final, post handling, concentration from the initially compounded amount as represented by the fresh standard solution. As computed, positive values imply that an increase in concentration has occurred within the returned sample, while negative values indicate sample loss. Values approximating zero infer no change.

As day to day fluctuations are known to influence chromatographic responses for duplicate sample injections, data analysis was limited to experiments performed on the same day under similar conditions. The average response factors of bracketing standard solution injections was determined, and individual samples compared directly with this value. This procedure was useful for normalizing the effects of instrumentation variability.

Experimental results were analyzed according to a standard statistical technique known as the Analysis of Variance (ANOVA). The basic assumption implicit in utilizing this technique is that the experimental observations (i.e., the deviations of the stored sample from the standard) are random variables, with well-defined distributional properties, that can be expressed as an additive linear function of the parameters that have been controlled in the experiment. If the appropriate assumptions are satisfied, it is then possible to test hypotheses regarding the effect of the individual parameters, or factors, that have been varied over two or more levels. If there is reason to believe that the necessary assumptions do not apply for the data, alternative analytical methods could be utilized; e.g., transforming the data, using non-parametric test methods, and/or performing an Analysis of Co-variance in which the standard calibration value is the co-variate and the observed peak area response the dependent variate. These techniques are described in detail in most statistical methods texts (11).

III. EXPERIMENTAL RESULTS

Results obtained from this three-set stability review are presented in Tables 3 through 5. The tabulated values reflect percentage variation of the stored samples from the expected sample content, based upon the initial formulation criteria. As such, values near zero imply best handling practice implementation, while negative values infer species loss and positive values imply an increase in concentration or, more likely, contamination. Deviation on either side of ideal is viewed as equally bad, as corrective practices would need to be implemented in either case.

Generally, the variability attributed to the analysis itself has been found to be small. Table 6 reflects the average peak area response, standard deviation and coefficient of variation for each compound within the standard preparation, as determined by repetitive injections throughout the individual analysis sequences. In addition, the 95 percent confidence levels surrounding the calculated means are presented.

As mentioned, the samples of Set III (sorbent resin set) consisted of the model compounds supported on XAD-2 solid sorbent resin; a combination which is unsuitable for direct GC analysis. In order to analyze these samples, it was necessary to extract them within a continuous Soxhlet extraction apparatus. The resulting solutions were then adjusted to the appropriate final volume and directly analyzed by GC.

Since the extraction process could possibly have interfered with the final results obtained, six non-stored samples were prepared and analyzed simultaneously with the stored samples to estimate the effect of this step. The results obtained are given in Table 7.

Observed average sample recoveries from the extraction process are found to range from 91.4 to 98.4 percent. However, as several of the listed data groups exhibit large variability around the computed average, no attempt has been made to adjust the results of Set III samples to account for the extraction process effects. Instead, tabulated values include all factors involved in sample storage, handling, and extraction.

Generally the data show that most compounds are recovered within $\pm 10\%$ of their original concentration for all three sets of conditions. The results are reviewed and analyzed in detail in Section IV.

Interpretation of the results obtained during Sets II and III analyses of n-Hexadecane concentration was difficult, due to the inordinately high recoveries which can not be assigned to any single parameter. Within Set II analyses, these variances, ranging from 10 to 65 percent above the formulated concentrations, were initially felt to arise from contamination. Observations made during analysis

TABLE 3

Analytical Results from Stored Probe Wash Samples (Set I)
(Percentage Deviation from Expected)

Run No.	Sample Identification ① ② ③ ④ *	Phenol	N-Methyl-aniline	Acenaphthene	n-Hexadecane	4,4'-Dichloro-biphenyl
1	AG-AA-H-RD	14.3	24.1	3.6	10.2	3.6
2	-AA-R-RD	4.9	0.5	-1.6	5.3	-6.3
3	-AA-C-RD	0.0	6.4	0.0	5.0	1.5
4	AG-AA-H-RL	-8.2	-0.3	-2.3	0.4	-8.5
5	-AA-R-RL	-0.5	5.7	3.8	6.3	7.2
6	-AA-C-RL	-1.6	3.6	1.2	3.7	3.6
7	AG-AA-H-CD	-6.0	5.5	-1.4	3.8	-2.7
8	-AA-R-CD	-1.9	13.7	-0.4	5.5	1.0
9	-AA-C-CD	-2.3	2.4	-2.1	2.7	0.6
10	AG-AN-H-RD	9.6	4.0	2.9	8.0	2.8
11	-AN-R-RD	-15.5	-0.7	-6.0	-2.2	-6.7
12	-AN-C-RD	-3.2	1.9	-2.7	1.4	11.0
13	AG-NN-H-RD	-0.4	5.2	1.8	6.0	0.0
14	-NN-R-RD	-2.1	1.0	-1.4	3.4	-0.5
15	-NN-C-RD	-4.2	0.8	-1.2	1.7	-1.5
16	AG-AN-H-RL	-8.2	-0.8	-2.3	-0.4	-8.5
17	-AN-R-RL	-7.6	-0.5	-1.9	-2.9	-7.6
18	-AN-C-RL	-13.7	-3.8	5.4	7.7	8.7
19	AG-NN-H-RL	0.0	3.1	2.7	5.3	6.7
20	-NN-R-RL	3.2	7.6	5.8	8.6	8.2
21	-NN-C-RL	2.6	8.3	5.8	9.8	9.2
22	AG-AN-H-CD	-4.1	0.7	-3.2	1.8	-1.4
23	-AN-R-CD	-17.6	-8.8	1.2	5.6	2.1
24	-AN-C-CD	-16.1	-10.0	0.8	5.6	2.1
25	AG-NN-H-CD	2.3	-0.3	3.1	7.7	2.0
26	-NN-R-CD	2.0	4.9	2.8	8.1	1.3
27	-NN-C-CD	-26.3	7.7	3.6	8.3	3.2

* Denotes factor combination observed during experiment (see Table 1).

continued....

Table 3 (continued)

Analytical Results from Stored Probe Wash Samples (Set I)
(Percent Deviation from Expected)

Run No.	Sample Identification ① ② ③ ④ *	Phenol	N-Methyl-aniline	Acenaphthene	n-Hexadecane	4,4'-Dichloro-biphenyl
28	P-AA-H-RD	2.2	6.6	4.7	-4.2	6.1
29	-AA-R-RD	1.8	4.0	2.6	-5.0	0.0
30	-AA-C-RD	-17.6	6.1	-2.3	-7.2	-4.6
31	P-AA-H-RL	4.7	3.4	5.7	-2.7	1.7
32	-AA-R-RL	9.8	-4.5	5.1	0.0	-1.6
33	-AA-C-RL	-16.2	-10.7	4.3	-0.2	1.1
34	P-AA-H-CD	-14.7	11.4	0.3	-4.3	-1.0
35	-AA-R-CD	31.6	18.9	3.2	-1.4	-0.6
36	-AA-C-CD	2.9	7.4	4.4	2.5	5.6
37	P-AN-H-RD	7.6	10.5	6.7	-1.2	7.8
38	-AN-R-RD	17.2	27.8	9.5	3.5	11.4
39	-AN-C-RD	0.4	6.8	2.5	-2.2	3.9
40	P-NN-H-RD	1.7	6.4	4.6	-3.6	1.6
41	-NN-R-RD	15.5	24.4	7.7	-0.7	3.4
42	-NN-C-RD	-7.9	5.1	0.3	-2.4	-2.9
43	P-AN-H-RL	12.7	1.1	4.0	-0.7	-2.0
44	-AN-R-RL	6.4	2.5	8.3	3.7	-15.6
45	-AN-C-RL	8.7	-4.5	4.0	0.7	7.5
46	P-NN-H-RL	12.7	-2.6	6.0	-2.2	4.4
47	-NN-R-RL	11.7	-4.2	4.0	-0.7	-1.6
48	-NN-C-RL	6.8	-5.6	3.6	1.5	10.2
49	P-AN-H-CD	-2.0	1.7	-1.3	-6.3	-2.0
50	-AN-R-CD	1.2	16.2	-4.3	-9.2	-5.6
51	-AN-C-CD	-8.3	8.0	3.6	-0.5	1.1
52	P-NN-H-CD	-16.8	-8.3	4.7	-2.5	-1.7
53	-NN-R-CD	-15.5	2.0	-10.2	-12.5	-15.7
54	-NN-C-CD	-9.1	7.0	-0.7	-2.5	-7.0

* Denotes factor combination observed during experiment (see Table 1).

Table 4

Analytical Results from Stored Sorbent Trap Condensate Extract Samples (Set II)
(Percent Deviation from Expected)

Run No.	Sample Identification+ ①②③④⑤*	Phenol	N-Methyl aniline	4-Chloro-benzaldehyde	Acenaph-thene	n-Hexa-decane	4,4'-Dichloro-biphenyl
1	P-NN-C-CD-N	0.8	2.6	3.0	-0.7	0.9	2.9
2	P-AN-C-RL-S	7.6	-6.0	7.1	2.3	1.9	3.9
3	P-AN-C-RL-S	8.4	-16.1	8.9	7.4	35.6	11.6
4	AG-AN-H-RD-N	2.9	21.6	0.7	2.5	0.9	-3.4
5	P-AA-H-CD-S	3.0	12.8	9.0	0.7	53.7	2.4
6	AG-AA-R-RL-N	-6.2	7.1	-0.7	-4.4	19.6	4.2
7	AG-NN-R-RD-S	-0.8	15.3	10.1	3.6	25.4	4.2
8	AG-AA-H-CD-S	-3.4	12.5	2.1	-1.6	22.8	12.0
9	AG-NN-H-RL-S	-2.3	15.2	3.1	0.9	23.3	-5.0
10	P-AA-C-RD-S	-0.3	8.6	5.2	0.0	42.0	-5.8
11	P-AN-R-CD-S	0.0	10.2	5.2	0.3	43.9	7.4
12	P-NN-H-RL-S	5.0	-10.9	13.0	3.4	65.2	10.5
13	P-NN-H-RL-S	6.0	-12.2	2.5	0.7	64.2	-0.5
14	AG-AA-C-RD-S	-6.8	6.8	0.0	-8.2	14.9	-1.0
15	AG-NN-C-CD-N	0.0	4.5	0.7	-8.0	14.2	0.0
16	P-NN-R-RD-S	-0.3	12.9	6.0	0.3	18.6	8.3
17	P-AN-H-RD-N	0.0	15.2	6.8	0.3	18.9	13.2
18	AG-AN-R-CD-S	-6.3	6.9	1.5	-6.7	16.6	8.7
19	P-NN-R-RD-S	1.9	13.5	6.0	1.3	24.2	2.9
20	P-NN-C-CD-N	-3.3	10.6	2.3	-4.4	14.1	4.9
21	P-AN-R-CD-S	0.8	5.4	5.4	-2.0	42.8	4.8
22	P-AA-C-RD-S	3.6	8.5	1.4	-0.7	46.3	5.8
23	P-AA-H-CD-S	2.6	12.5	7.6	0.7	20.1	11.1
24	P-AA-R-RL-N	-1.3	-2.3	3.3	-3.3	15.2	7.3
25	P-AA-R-RL-N	-1.0	3.1	-1.5	-4.7	14.4	-0.5
26	P-AN-H-RD-N	2.9	17.5	14.3	3.8	21.5	6.8
27	AG-AN-C-RL-S	-3.1	9.1	5.3	-4.0	20.3	-1.9

+ Equivalent sample identification codes indicate separate preparations of duplicate samples. Presented in randomized order as performed in experiment.

* Denotes factor combination observed during experiment (see Table 1).

TABLE 5

Analytical Results from Stored Sorbent Resin Samples (Set III)
(Percent deviation from expected)

Run No.	Sample Identification+ ① ② ③ ④*	Phenol	N-Methyl-aniline	4-Chloro benzaldehyde	Acenaph-thene	n-Hexa-decane	4,4'-Dichloro-biphenyl
1	AG-NN-R-RD	0.0	-3.4	-4.0	-2.4	5.2	-1.4
2	AG-AA-C-CD	3.1	1.3	-2.1	-1.0	24.2	-0.3
3	AG-AA-H-CD	-0.5	2.9	-1.2	0.0	19.7	1.2
4	AG-NN-H-RD	1.1	-9.0	-6.1	-6.9	11.7	-8.2
5	AG-NN-C-CD	4.3	2.9	-4.0	-3.7	15.1	-5.1
6	AG-AA-R-CD	-9.9	-2.1	-6.1	-5.6	15.3	1.0
7	AG-NN-R-CD	1.8	8.1	-0.7	-1.2	15.5	0.3
8	AG-NN-H-CD	1.8	1.3	-3.0	0.8	19.0	-1.4
9	AG-AA-R-RD	0.0	1.3	-1.5	0.8	18.6	-2.1
10	AG-NN-C-RD	6.0	2.7	-3.0	-1.3	17.1	3.1
11	AG-AA-H-RD	-0.6	5.0	-0.8	0.0	18.4	-1.1
12	AG-AA-C-RD	1.2	6.8	-0.8	-0.5	18.8	-2.2
13	AG-AA-H-CD	0.6	3.6	-3.1	-3.2	15.9	-4.9
14	AG-NN-R-CD	3.5	-6.0	-0.8	-1.1	18.4	-2.6
15	AG-NN-C-RD	-4.1	1.6	-4.9	-2.2	17.0	-3.7
16	AG-AA-C-CD	-2.9	6.3	-4.2	-0.9	17.9	-1.3
17	AG-AA-R-RD	3.9	2.8	-8.8	-0.7	19.7	-1.0
18	AG-NN-H-RD	4.9	6.3	-11.2	-1.4	17.5	-2.0

+ Equivalent sample identification codes indicate separate preparation of duplicate samples.

* Denotes factor combination observed during experiment (see Table 1).

Table 6

Statistical Summary of Standard Calibration Samples

		Phenol	N-Methyl-aniline	4-Chloro-benzeldehyde	Acenaphthene	n-Hexadecane	4,4'-Dichloro-biphenyl
Set I	Mean	274	356		265	504	200
Probe Wash	st'd dev.	36.9	27.3	not	9.7	20.1	21.8
Series	coef. of var.	13.5%	7.7%	analyzed*	3.7%	4.0%	10.9%
	95%						
50 Observations	confidence limits	263.5 - 284.5	348.2 - 363.8		262.2 - 267.8	498.3 - 509.7	193.8 - 206.2
Set II	Mean	376	366	261	295	456	236
Condensate	st'd dev.	9.4	8.3	9.8	10.9	15.1	19.4
Extract	coef. of var.	2.5%	2.3%	3.8%	3.7%	3.3%	8.2%
Series							
	95%						
12 Observations	confidence limits	370.1 - 381.9	360.8 - 371.2	254.8 - 267.2	288.2 - 301.8	446.5 - 465.5	223.8 - 248.2
Set III	Mean	15.6	18.5	16.0	42.0	21.9	29.6
Sorbent Resin	st'd dev.	1.1	1.4	0.9	2.6	1.7	1.8
Series	coef. of var.	7.1%	7.6%	5.6%	6.2%	7.8%	6.1%
	95%						
16 Observations	confidence limits	15.0 - 16.2	17.8 - 19.2	15.5 - 16.5	40.6 - 43.4	21.0 - 22.8	28.6 - 30.6

* This species was not present in Set I samples.

TABLE 7

Results of Extraction Step Experiment

(Percentage Deviations from Unspiked Standard)

Standard Spikes	Phenol	N-Methyl Aniline	4-Chloro-Benzaldehyde	Acenaphthene	n-Hexa-decane	4,4'-Dichloro-biphenyl
Run No. 1	-5.6	-0.5	-8.5	-1.4	-1.4	-1.7
2	-1.9	+3.4	-3.6	-2.0	-1.3	-4.1
3	-8.6	+3.9	-4.2	-2.3	-1.3	-4.5
4	-14.1	-12.5	-7.8	-2.1	-2.3	-2.0
5	-9.0	-3.7	-13.7	-6.4	-2.3	-1.3
6	-3.0	-5.3	-13.7	-0.7	-0.9	-0.7
Average	-7.03	-2.45	-8.58	-2.48	-1.58	-2.38
95% Confidence Interval Limits	-2.3 to -11.7	-8.9 to +4.0	-4.0 to -13.6	-0.4 to -4.6	-1.0 to -2.2	-0.7 to -4.0

tended to support this theory, as a shoulder was noted on the chromatogram part way through the hexadecane peak. In addition, where a relatively smooth baseline had once existed, several smaller but reproducible peaks were in evidence. To determine the contaminants' composition, two representative samples of the condensate extract set were studied by capillary gas chromatography-mass spectrometry (GC-MS). The chromatograms obtained from these analyses are presented in Figures 1 and 2.

Referring to Figure 1, it is clear that a homologous series of olefins is present within the retained sample (P-NN-H-RL-S), and that the species identified as a C_{16} olefin is the probable cause of much of the observed extra hexadecane content (65%). As olefins are major constituents of both mold release compounds and polyethylene bottles, it is likely that the contaminant series results from the extraction of container walls by the methylene chloride.

However, the excessive n-hexadecane content observed in most of the samples of Set II and Set III cannot be singly attributed to the extraction of contaminants from the polyethylene sample bottles. With reference to Figure 2, a chromatogram of a second sample (P-NN-R-RD-S) from Set II, where a 25 percent excess of n-hexadecane had been reported, indicates that even though the same homologous series of contaminants is present, their apparent contribution to the reported excess is not large (even on a proportional basis). Furthermore, reference to the results of Set I analyses, indicates that not all samples stored in polyethylene bottles of the same lot and under similar conditions will become contaminated.

Comparably, review of a typical sample from the serbent resin set (see Figure 3) shows that no olefin species are contained within a sample exhibiting a net gain of 24 percent n-hexadecane. As all the tested resin combinations in this set, and many samples in Set II as well, were stored in amber glass bottles, the extraction of olefins from the container walls is not considered likely.

Thus, the origin and nature of all factors influencing the n-hexadecane concentration within Set II and Set III samples is not completely clear. While it is obvious that some of the samples have become contaminated with components extracted from the walls of the polyethylene sample bottles, this alone cannot account for all of the encounted hexadecane excesses. Other factors, including possibly inaccurate sample formulation, must be considered as having influenced some of the reported results, making the interpretation of Set II and III n-hexadecane results impossible.

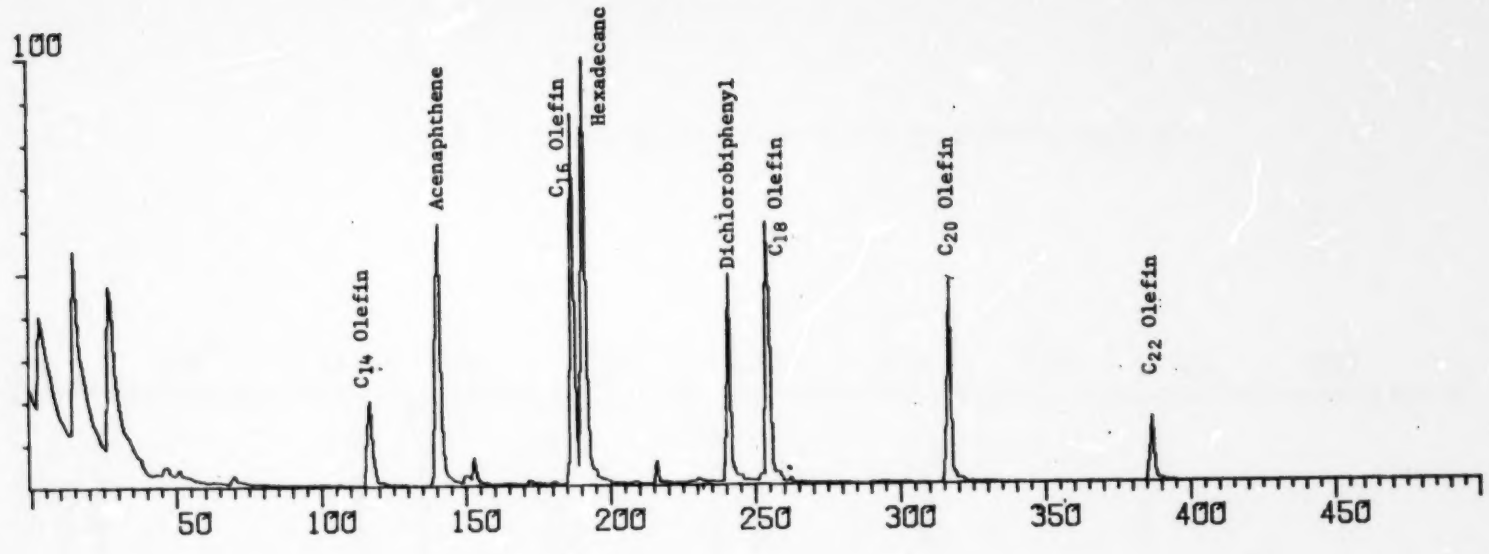


Figure 1. GC/MS Chromatogram of Set II Sample P-NN-H-RL-S

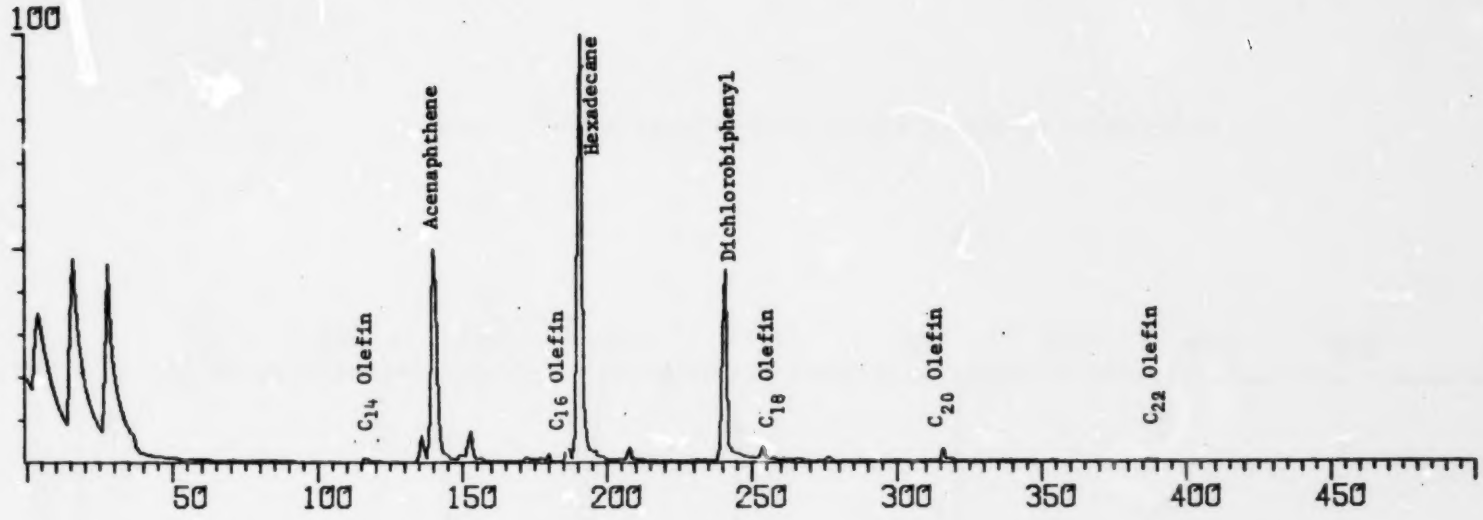


Figure 2. GC/MS Chromatogram of Set II Sample P-NN-R-RD-S

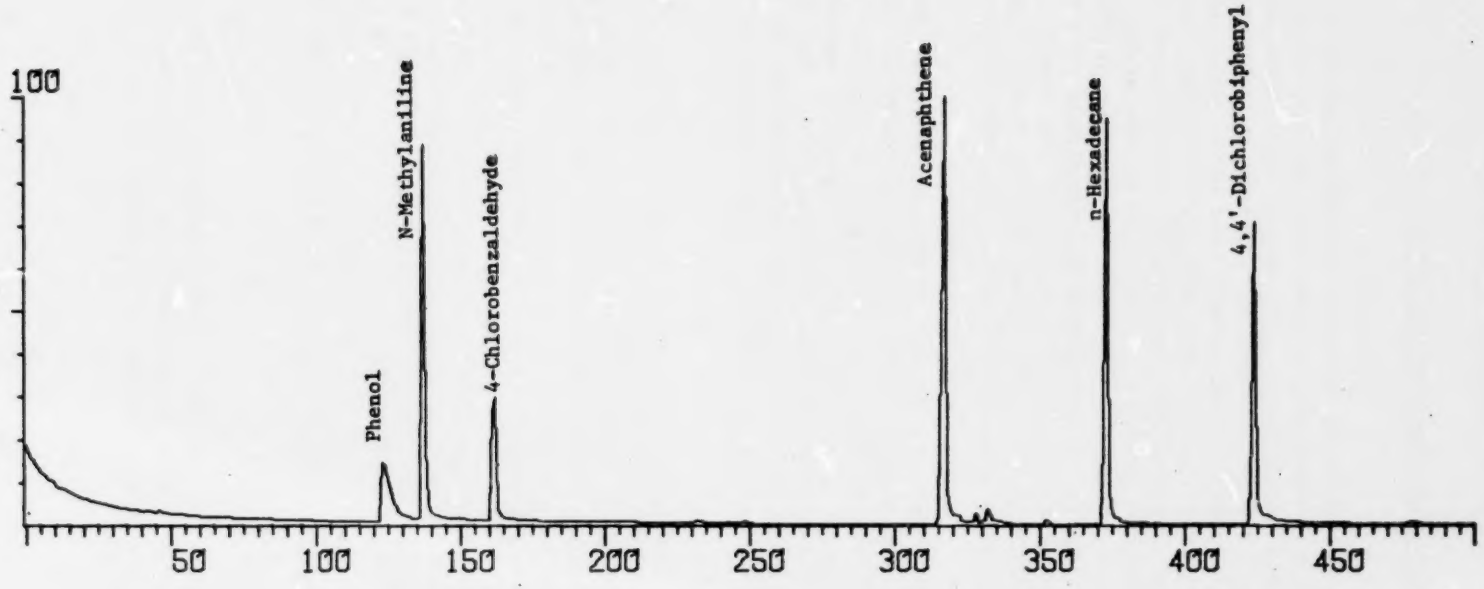


Figure 3. GC/MS Chromatogram of Set III Sample AG-AA-C-CD

IV. DISCUSSION OF RESULTS

A. Statistical Analysis of Experimental Data

The changes in composition (percent deviation) of each compound, given in Tables 3-5, are observed to vary over the set of experimental input conditions considered. The statistical technique ANOVA provided a mechanism to allocate this variability in the data to identifiable sources; namely, those conditions which were controlled in the design of the experiment (i.e., type of container, head space composition, shipment temperature, and storage condition). Furthermore, the ANOVA technique provides an estimate of the normal or chance variation in the data that has been an inherent part of performing the experiment. By comparing the magnitude of the difference attributable to varying each factor over the levels tested to the normal variation in the data, those factors having a measurable (i.e., statistically significant) effect can be identified.

To illustrate this concept, an ANOVA table is given in Table 8 for the set of 54 phenol observations reported in Table 3. Although the basic experiment has not been replicated, it is possible to estimate chance variation (i.e., experimental error) by assuming all three and four factor interactions are negligible. For the phenol data, the experimental error variance is estimated to be 82.81: thus the standard error is $\sqrt{82.81} = \pm 9.1$ percent on a per observation basis.

The ANOVA table indicates those effects and two-factor interactions that appear to contribute significantly to the variability of the data. To illustrate this concept, the average values of each level of each factor are given in Table 9. For example, the significant contribution attributable to varying types of container on the phenol data can be explained by the fact that amber glass, on the average, yielded a loss of 3.7 percent, while the 27 experiments using polyethylene yielded a positive deviation of 1.8 percent phenol. This difference of 5.5 percent is statistically significant, since the ANOVA table provides the data to calculate a 95 percent confidence interval about this difference:

$$\text{Estimated "container effect"} = (-3.7 - 1.8) \pm (2.048)\sqrt{\frac{2(82.81)}{27}}$$

$$= -5.5\% \pm 5.1\%$$

Therefore, this experiment indicates the likelihood that the use of polyethylene results in phenol measurements of 0.4 percent to 10.6 percent higher than would be observed using amber glass, a positive (non-zero) contribution.

TABLE 8

Analysis of Variance

Probe Wash Experiment (Phenol)

Source of Variation	Degrees of Freedom	Sum of Squares	Mean Square	F
Main Effects				
Container	1	406.3	406.3	4.91*
Head Space Composition	2	38.6	19.3	< 1
Shipment Temperature	2	675.2	337.6	4.08*
Storage Condition	2	574.3	287.1	3.47*
Interactions				
Container x Head Space Composition	2	426.4	213.2	2.57
Container x Shipment Temperature	2	363.5	181.7	2.19
Container x Storage Condition	2	159.6	79.8	< 1
Head Space Composition x Shipment Temperature	4	327.9	82.0	< 1
Head Space Composition x Storage Condition	4	689.1	172.3	2.08
Shipment Temperature x Storage Condition	4	175.7	43.9	< 1
Error	28	2318.8	82.8	
Total	53	6155.5		

Standard Error per sample = $\sqrt{82.81}$ = $\pm$ 9.1%

* Indicates significance at 5% test level

TABLE 9

Probe Wash Experiment (Set I)
(Average Percentage Deviation from Expected)

Factor	Level	No. of Observations	Phenol	N-Methyl aniline	Acenaphthene	n-Hexadecane	4,4'-Dichlorobiphenyl
Container	Amber Glass	27	-3.7	3.0	0.7	4.7	1.2
	Polyethylene	27	1.8 *	5.1	3.0 *	-2.2 **	0.1
Head Space Composition	Air/Air	18	0.2	5.8	1.6	1.1	0.4
	Air/Nitrogen	18	-1.8	2.9	1.5	0.7	0.5
	Nitrogen/Nitrogen	18	-1.3	3.5	2.4	1.8	1.1
Shipment Temperature	Hot	18	0.4	4.0	2.2	0.8	0.5
	Room	18	2.5 *	6.1	1.6	0.9	-1.5 *
	Cold	18	-5.8	2.0	1.7	2.0	3.0
Storage Condition (Temperature/Light)	Room/Dark	18	1.3	7.8	1.8	0.9	1.7
	Room/Light	18	1.3 *	-0.1 **	3.5 *	2.1	1.3
	Cold/Dark	18	-5.6	4.4	0.2	0.7	-1.0
Average		54	-1.0	4.1	1.8	1.2	0.6
Standard Error Per Sample			9.1	6.3	3.4	3.5	4.9
Significant Interactions:			None	Cont. x Hd. Sp. Comp. * Cont. x Stor. Cond. *	None	None	Hd. Sp. Comp. x Stor. Cond. *

* Indicates significance at 5% test level.
** Indicates significance at 1% test level.

Similarly, all other significant differences are identified in Table 9. The Set I experiment was structured in such a way that it was possible to investigate not only the influence of each factor independently, but also the interactive or combined effect of all factor pairs. Therefore, significant two-factor interactions are also given in Table 9. The nature and interpretation of the two-factor interactions observed to be significant are demonstrated by the two-way tables and graphs given in Table 10 and Figure 4. For example, it is observed that n-methylaniline readings were approximately 2 percentage points higher with glass than with polyethylene when tested with air/air or nitrogen/nitrogen head space compositions. However, this situation was very different when air/nitrogen was used, in that glass yielded nearly 10 percent lower readings than polyethylene.

The condensate extract (Set II) experimental design was different from Set I in that all possible factor/level combinations were not tested. It had been possible as a result of analzying the data from Set I to eliminate some of the small effect combinations, The plan used, known as a Fractional Factorial design, required only twenty-seven unique test conditions in order to estimate each factor independently. Furthermore, only the two-factor interactions involving the container effect were estimable. This analysis required the assumption that all other two-factor and higher order interactions had a negligible effect on the data.

An illustrative example of the components of variation are given for phenol data in Table 11. It is observed that only the container effect is statistically significant; i.e., an increase between 1.4 and 7.4 percent can be expected in phenol measurements when polyethylene is used instead of amber glass.

Table 12 summarizes the results of all ANOVA's computed for Set II. Significant interactions are illustrated in tabular (Table 13) and graphic form (Figure 5) as described for Set I data. Similar analytical results are given for the Sorbent Resin (Set III) experiment in Tables 14 and 15.

B. Interpretation of Results

The overall purpose of this set of experiments has been to detect measureable differences in the recoveries of select model compounds that are directly attributable to the variation of specific shipment and storage procedures. Subsequent to detection, the extent of difference found would be directly compared with the accuracy requirements of a Level 1 survey analysis to determine if, in fact, the measured variability could interfere with the desired goals of such a survey.

As such, the laboratory experiments were designed and conducted in such a way that differences were likely to be detected if, indeed,

TABLE 10

Probe Wash Experiment (Set I)
Significant Two-factor Interactions

COMPOUND

N-Methylaniline:

Head Space Composition

Container	AA	AN	NN	
Glass	6.8	-2.0	4.3	*
Poly	4.7	7.8	2.7	

Storage Condition

Container	RD	RL	CD	
Glass	4.8	2.5	1.8	*
Poly	10.9	-2.8	7.1	

4,4'-Dichlorobiphenyl:

Storage Condition

Head Space Composition	RD	RL	CD	
AA	0.0	0.6	0.5	*
AN	5.0	-2.9	-0.6	
NN	0 0	6.2	-3.0	

* Indicates significance at 5% test level.

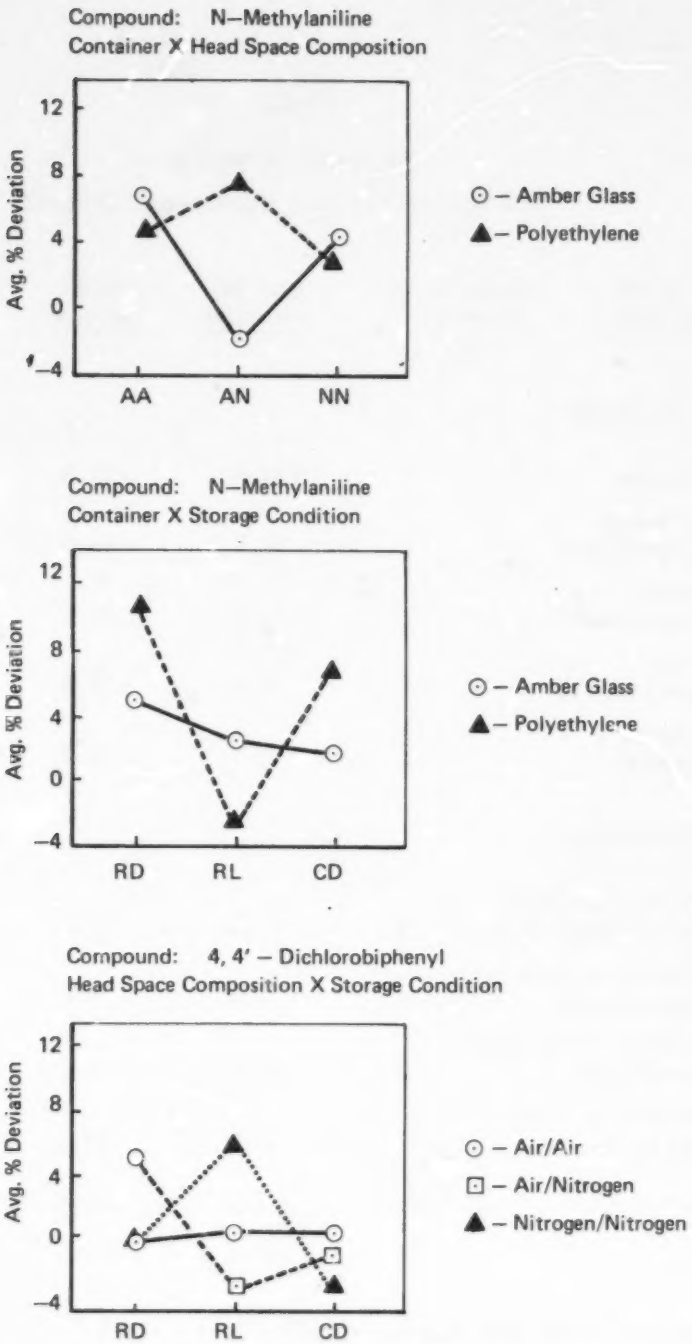


**FIGURE 4. PROBE WASH EXPERIMENT (SET I)
SIGNIFICANT TWO-FACTOR INTERACTIONS**

TABLE 11

Analysis of Variance

Condensate Extract Experiment (Phenol)

Source of Variation	Degrees of Freedom	Sum of Squares	Mean Square	F
Main Effects				
Container	1	113.8	113.8	10.16**
Head Space, Composition	2	31.0	15.5	1.40
Shipment Temperature	2	29.6	14.8	1.33
Storage Conditions	2	5.9	2.9	< 1
Catalytic Species	1	4.7	4.7	< 1
Interactions				
Container x Head Space Composition	2	34.0	17.0	1.53
Container x Shipment Temperature	2	12.9	6.4	< 1
Container x Storage Condition	2	12.6	6.3	< 1
Container x Catalytic Species	1	38.3	38.3	3.45
Error	11	122.1	11.1	
Total	26	405.0		

Standard Error per sample = $\sqrt{11.10}$ = 3.33%

** Indicates significance at the 1% test level.

TABLE 12

Condensate Extract Experiment (Set II)
(Average Percentage Deviation from Expected)

Factor	Level	No. of Observations	Phenol	N-Methylaniline	4-Chlorobenzaldehyde	Acenaphthene	n-Hexadecane	4,4'-Dichlorobiphenyl
Container	Amber Glass	9	-2.9 **	11.0 **	2.5 *	-2.9 **	17.6	2.0
	Polyethylene	18	1.5	4.8	5.9	0.3	30.2	5.4
Head Space Composition	Air/Air	9	-1.1	7.7	2.9	-2.4	27.7	3.9
	Air/Nitrogen	9	1.5	7.1	6.1	0.4	22.5	5.7
	Nitrogen/Nitrogen	9	-0.3	5.7	5.2	-0.3	27.8	3.1
Shipment Temperature	Hot	9	0.7	9.4	6.6	1.3	32.3	5.2
	Room	9	-1.5	8.0 *	3.9	-1.7 *	24.5	5.3
	Cold	9	0.8	3.2	3.8	-1.8	21.1	2.3
Storage Condition (Temperature/Light)	Room/Dark	9	0.3	13.3	5.6	0.3	23.6	3.4
	Room/Light	9	0.3	-1.4 **	4.6	-0.2	28.9	3.3
	Cold/Dark	9	-0.6	8.7	4.1	-2.4	25.5	6.0
Catalytic Species	Stainless Steel	18	0.3	5.8	5.5	-0.1	32.3 *	4.4
	None	9	-0.6	8.9	3.2	-2.1	13.3	3.9
Average Standard Error Per Sample		27	0.01	6.8	4.8	-0.8	26.0	4.3
			3.33	3.78	3.62	2.49	16.9	5.4
Significant Interaction			None	Cont. x Stor. Cond. **	None	Cont. x Ship. Temp. *	None	None

* Indicates significance at 5% test level.

** Indicates significance at 1% test level.

TABLE 13

Condensate Extract Experiment (Set II)
Significant Two-factor Interactions[+]

COMPOUND

N-Methylaniline:

Storage Condition

		RD	RL	CD	
Container	Glass	14.6	10.5	8.0	**
	Poly	12.7	-7.4	9.0	

Acenaphthene:

Shipment Temperature

		Hot	Room	Cold	
Container	Glass	0.6	-2.5	-6.7	*
	Poly	1.6	-1.3	0.6	

[+]Only those interactions involving the factor "Container" are estimable from this design.

*Indicates significance at 5% test level.
**Indicates significance at 1% test level.

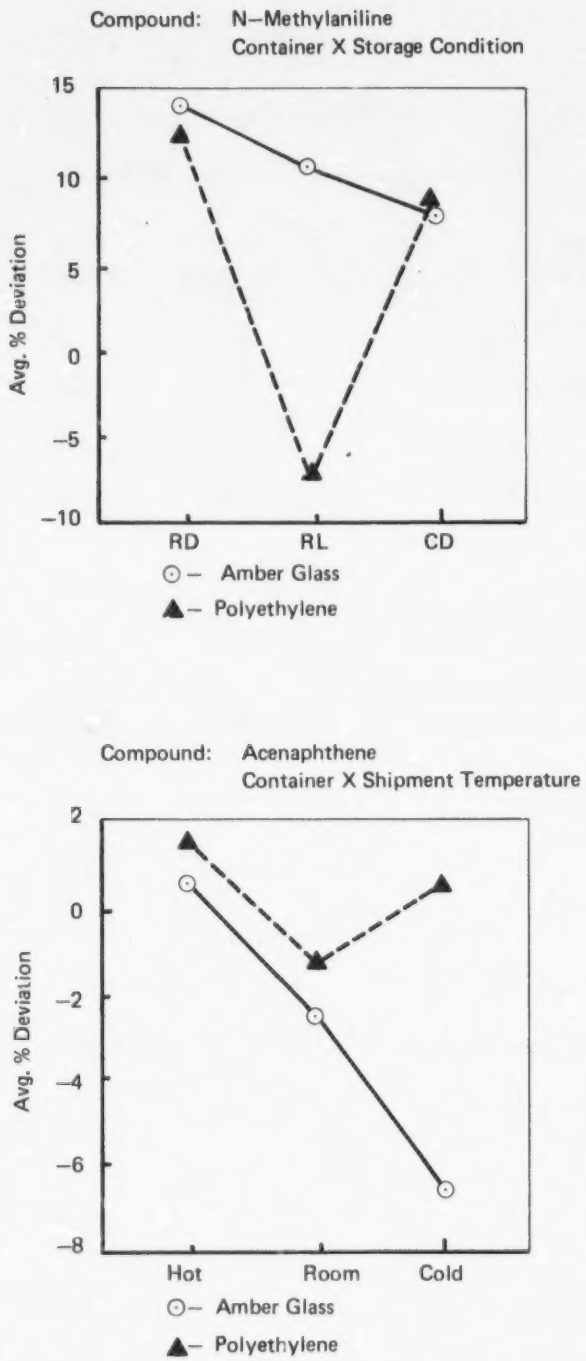


FIGURE 5. CONDENSATE EXTRACT EQUIPMENT (SET II) SIGNIFICANT TWO-FACTOR INTERACTIONS

TABLE 14

Analysis of Variance

Sorbent Resin Experiment (Phenol)

Source of Variation	Degrees of Freedom	Sum of Squares	Mean Square	F
Main Effects				
Head Space	1	33.1	33.1	1.64 (non-sig.)
Shipment Temperature	2	7.4	3.7	<1
Storage Condition	1	6.2	6.2	<1
Interactions				
Head Space Composition x Shipment Temperature	2	3.5	1.8	<1
Head Space Composition x Storage Condition	1	17.9	17.9	<1
Shipment Temperature Storage Condition	2	8.2	4.1	<1
Error	8	161.4	20.2	
Total	17	237.7		

Standard Error per sample = $\sqrt{20.2}$ = ±4.5%

TABLE 15

Sorbent Resin Experiment (Set III)
(Average Percentage Deviation from Expected)

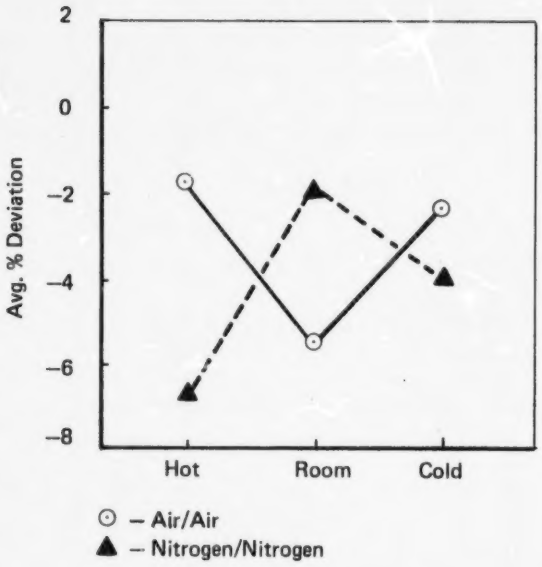
Factor	Level	No. of Observations	Phenol	N-Methyl-aniline	4-Chloro-benzalde-hyde	Acenaph-thene	n-Hexa-decane	4,4'-Dichloro-biphenyl
Head Space Composition	Air/Air	9	-0.6	3.1	-3.2	-1.2	18.7	-1.2
	Nitrogen/Nitro-gen	9	2.1	0.5	-4.2	-2.2	15.2	-2.3
Shipment Temperature	Hot	6	1.2	1.7	-4.2	-1.8	17.0	-2.7
	Room	6	-0.1	0.1	-3.6	-1.7	15.4	-1.0
	Cold	6	1.3	3.6	-3.2	-1.6	18.4	-1.6
Storage Condition (Temp/Light)	Room/Dark	9	1.4	1.6	-4.6	-1.6	16.0	-2.1
	Cold/Dark	9	0.2	2.0	-2.8	-1.8	17.9	-1.5
Average		18	0.8	1.8	-3.7	-1.7	16.9	-1.8
Standard Error Per Sample			4.5	5.6	2.1	2.2	4.3	3.3
Significant Interactions			None	None	Hd. Sp. Comp. x Ship. Temp. *	None	None	None

* Indicates significance at the 5% test level.

Compound: 4—Chlorobenzaldehyde

		Shipment Temperature			
		Hot	Room	Cold	
Head Space Composition	Air	−1.7	−5.5	−2.4	*
	Nitrogen	−6.8	−1.8	−4.0	

Head Space Composition X Shipment Temperature



* Indicates significance at the 5% test level.

FIGURE 6. SORBENT RESIN EXPERIMENT (SET III) SIGNIFICANT TWO-FACTOR INTERACTION

they did exist (that is, the design was "powerful" in the statistical sense). Furthermore, it should be recognized that these experiments were intended to yield estimates of the magnitude of the true, but unknown, average effect caused by varying a parameter over two or more levels, and that these estimates were to be measured with reasonable analytical precision. Thus, as an example, it was found in Set I experiments that a sample stored in polyethylene can be expected to yield a larger phenol concentration than comparable samples stored within amber glass containers. Furthermore, the concentration of phenol in polyethylene can be expected to be from 0.4 to 10.6 percent higher on the average.

Although this suggests that such a difference is "real" in the sense that repetitions of this experiment would yield similar findings, the added contribution of even 10 percent is considered to be well within normally accepted analytical limits for Level 1 analyses.

With this in mind, it becomes clear that none of the tested combinations produce differences large enough to impact upon the results of a Level 1 survey analysis. Thus, any reasonable combination of shipment temperature, storage condition, container, head space composition and catalytic species content can be used for a short period of time. Furthermore, inasmuch as a ± 10 percent range is normally acceptable for quantitative chemical analysis of samples, it appears that most of these same conditions can be directly applied to quantitative analysis routines. However, the tabulated data also suggest that some caution should be exercised before these conditions are unilaterally applied.

One occasion implying caution, occurs during the analysis of the n-Hexadecane results in Set II experiments. As was previously mentioned, a homologous series of olefins apparently was extracted from several, but not all, of the polyethylene containers used during this experimental sequence. The C_{16} member of this series contributed directly to the observed excessive n-hexadecane concentrations, as it was integrated as part of the model compound GC response, causing an average increase of 26 percent to be found within the stored samples. However, the distribution of the olefin series was not uniform throughout all similarly handled samples implying that some of this observed variability was produced by other factors which can not be identified by the experimental conditions used. As the origin of all factors contributing to the excessive hexadecane content are unclear, judgment of the handling condition effect upon species similar to n-hexadecane must be reserved.

What is important from this experiment, however, is the fact that it emphasizes that polyethylene containers should not be used for the storage of samples in organic solvents. While other results of this study imply that plastic containers do not affect the model compounds recoveries (due to loss or concentration), this sequence of experiments shows that undesirable contaminants can arise during confinement within polyethylene and interfere with eventual analysis.

As there is no way to insure that spurious contaminants will not be extracted from the polyethylene, only those materials unlikely to react with the container should be retained in them.

A review of N-Methylamiline data from both Set I and Set II also is of interest. As listed, only those samples stored in diffuse sunlight and at room temperature exhibit a net decrease in the N-Methylaniline content. From Tables 9 and 12 it is noted that decreases of 0.1 and 1.4 percent are encountered while other tested storage conditions (room or cold temperature and in the dark) produce increases ranging from 4.0 to 14.0 percent of the intially formulated concentrations. The noted decreases are even more pronounced when a review of the storage condition x container composition interaction is made. As seen from Tables 10 and 13, those samples stored in polyethylene and exposed to diffuse sunlight lose between 2.5 and 7.5 percent of the initially compounded N-Methylaniline content, while all other tested conditions (including both polyethylene and amber glass containers stored in the dark, and amber glass stored in diffuse sunlight) show increased levels ranging from approximately 1.5 to 15.0 percent.

As N-Methylaniline is known to be photoreactive, it is presumed that the noted species loss is attributable to some amount of photooxidation. While the amber glass bottle appears to have adequately screened out the reactive ultraviolet light, the polyethylene did not. This is expected because polyethylene is transparent in the ultraviolet region and will allow some light to penetrate and degrade the aniline. The amber glass, on the other hand, absorbs the ultraviolet light prohibiting photodegradation of the amine. Once again this implies that the utilization of polyethylene should be restricted to those instances where it is known that stored samples will not degrade solid samples.

V. CONCLUSIONS AND RECOMMENDATIONS

Excluding the storage of certain materials within polyethylene containers or in the presence of diffuse sunlight, any reasonable combination of handling procedures appears to be suitable for the short term storage of Level 1 survey analysis samples. As Level 1 procedures only require the identification of sample constituents within a range of ±2 or 3 times of what actually exists, the relatively small perturbations encountered during this experimental review will not detract from the value of the accumulated data. In fact, based on these results, it appears that several of the tested conditions, including shipment temperatures (below 38°C), storage conditions (storage at either room or reduced temperature and in the dark) head space compositions (air or nitrogen) and amber glass containers, are appropriate for the storage of quantitative samples where variation must be held to ±10 percent.

The utilization of polyethylene sample bottles for storage of organic materials is not appropriate, as it has been demonstrated during Set II experiments that spurious contaminants can be extracted even from rigorously precleaned containers. Furthermore, polyethylene is not suitable for the storage of photo-reactive substances. As noted from Sets I and II, N-Methylaniline results, samples stored for short periods of time in sunlight (even indirect) can be lost through photo-degradation. While amber glass bottles absorb the harmful ultraviolet wavelengths, polyethylene, which is transparent, allows the light to penetrate. Therefore, it is recommended that these containers only be used for the storage of samples that are known not to degrade.

Furthermore, it is recommended that attempts be made to minimize the contact of organics species with catalytic agents. Although the presence of acid extractable stainless steel constituents has not been proven to be significant during this analysis, the effect of these and other catalytic species has not been fully explored.

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APPENDICES

APPENDIX A - Formulation Procedures for Samples

TABLE A1 - Formulation Procedure for Probe Wash Samples (Set I)

TABLE A2 - Formulation Procedure for Sorbent Trap Condensate Extract Samples (Set II)

TABLE A3 - Formulation Procedure for Sorbent Resin Samples (Set III)

TABLE A1

Formulation Procedure for Probe Wash Samples (Set I)

Amber Glass Samples - 27 samples of 300 mls each

9 liters total volume of a 1:1 mix of methylene chloride:methanol

	conc µg/ml
Phenol	168
N-Methylaniline	167
Acenaphthene	95
n-Hexadecane	166
4-4'-Dichlorobiphenyl	103

Polyethylene Sample - 27 samples of 300 mls each.

9 liters total volume of a 1:1 mix of methylene chloride:methanol

	conc µg/ml
Phenol	155
N-Methylaniline	162
Acenaphthene	103
n-Hexadecane	164
4,4'-Dichlorobiphenyl	94

TABLE A2

Formulation Procedure for Sorbent Trap Condensate Extract Samples (Set II)

White liquor extract - unspiked - 9 samples of 300 mls each.

White liquot - 3 liters of an 0.05 M HCl and 0.005 M H_2SO_4 in distilled water prepared (metallic content analyzed by ICPOES) and then extracted with an equal volume of methylene chloride. Methylene chloride extract retained as white liquor extract. To this, the following concentrations of model compounds was added.

	µg/ml
Phenol	165
N-Methylaniline	169
4-Chlorobenzaldehyde	166
Acenaphthene	99
n-Hexadecane	168
4,4'-Dichlorobiphenyl	100

Green liquor extract - spiked - 18 samples of 300 mls each.

Green liquor - A piece of 316 stainless steel placed in 100 mls of 1.0 M HCl and stirred for 3 hours, producing green acidic solution.

A 6-liter volume of 0.05 M HCl, 0.005 M H_2SO_4 and distilled water was prepared using the concentrate as a base (metallic content analyzed by ICPOES). The total volume was then extracted with an equal amount of methylene chloride. Methylene chloride (green liquor extract) retained and metallic content analyzed. (See Appendix B). Aqueous phase discarded.

	µg/ml
Phenol	165
N-Methylaniline	169
4-Chlorobenzaldehyde	167
Acenaphthene	101
n-Hexadecane	168
4,4'-Dichlorobiphenyl	101

TABLE A3

Formulation Procedure for Sorbent Resin Samples (Set III)

Amber Glass bottles - 24 samples

Stock solution - seven model compounds added to 200 mls pentane to produce following concentrations:

	µg/ml
Phenol	66
N-Methylaniline	106
4,-Chlorobenzaldehyde	60
Acenaphthene	99
n-Hexadecane	64
4,4'-Dichlorobiphenyl	100

Five mls of stock solution pipetted onto 15± 1 gm of XAD-2 resin contained in amber glass bottles. After handling period, samples were removed and extracted (Soxhlet) with methylene chloride for 24 hours. Extract volume brought to 250 mls and analyzed.

APPENDIX B - Analytical Results of Metallic Content Contained Within Green Liquor, White Liquor and Green Liquor Extract

TABLE B1 - Metallic Content of Green and White Liquors As Determined by Inductively Coupled Plasma Optical Emission Spectrometry (ICPOES)

TABLE B2 - Metallic Content of White Liquor, Green Liquor and Green Liquor Extract by Emission Spectrometry As Determined by Spark Emission Spectrometry

TABLE B1

METALLIC CONTENT OF GREEN AND WHITE LIQUORS BY ICPOES

(Values in µg/ml)

Compound / Element	White Liquor	Green Liquor	Detection Limits (in H_2O)
Li	ND	ND	0.3
Be	ND	ND	0.001
B	ND	ND	0.05
Na	ND	0.9	0.3
Mg	ND	0.01	0.006
Al	ND	0.11	0.05
Si	0.01	0.5	0.01
K	ND	ND	1
Ca	ND	0.15	0.001
Ti	ND	0.003	0.002
V	ND	0.009	0.003
Cr	ND	2.3	0.005
Mn	0.004	0.22	0.001
Fe	ND	8.3	0.003
Co	0.13	0.005	0.005
Ni	ND	1.6	0.01
Cu	0.08	0.14	0.01
Zn	ND	0.13	0.008
Sr	ND	0.008	0.002
Mo	0.02	0.31	0.01
Ag	ND	ND	0.01
Cd	ND	ND	0.001
Sn	ND	ND	0.01
Ba	ND	0.003	0.001
Pb	ND	ND	0.01

ND = not detected

TABLE B2

METALLIC CONTENT OF WHITE LIQUOR, GREEN LIQUOR AND GREEN LIQUOR EXTRACT BY EMISSION SPECTROMETRY

(Values in µg/ml)

Compound / Element	White Liquor	Green Liquor	Green Liquor Extract	Detection Limits
Li	ND	ND	ND	0.1
Be	ND	ND	ND	0.001
B	ND	ND	ND	0.005
Na	0.5	3	0.05	0.05
Mg	0.005	0.01	0.001	0.001
Al	ND	0.05	ND	0.01
Si	0.01	0.5	0.005	0.01
K	ND	ND	ND	10
Ca	0.01	0.01	ND	0.005
Ti	ND	ND	ND	0.01
V	ND	ND	ND	0.01
Cr	ND	4	ND	0.01
Mn	ND	0.8	ND	0.005
Fe	0.01	10	ND	0.01
Co	ND	ND	ND	0.05
Ni	ND	3	ND	0.05
Cu	0.05	0.10	ND	0.001
Zn	ND	ND	ND	0.05
Sr	ND	0.1	ND	0.01
Mo	ND	0.1	ND	0.01
Ag	0.001	0.005	0.001	0.001
Cd	ND	ND	ND	0.01
Sn	ND	ND	ND	0.05
Ba	ND	ND	ND	0.1
Pb	ND	ND	ND	0.01

ND - Not detected

APPENDIX C - Gas Chromatography Conditions Employed During Analysis

TABLE C1

Gas chromatographic conditions:

Instrument: Varian Assoc. Model 2700 with FID

Column: 6' x 1/8" SS containing 10% OV - 101 on 100/120 Supelcoport

Conditions:

He 30 ml/min

H_2 ∿30 ml/min flows adjusted slightly to optimize performance

air ∿300 ml/min

injection port 260°-270°C

detector 300°-310°C

Program: 50° (5 min) $\xrightarrow{10°/min}$ 215° (4 min)

Calculations: Spectra-Physics System I Computing Integrator